INTRODUCTION

Currently, a novel oral colon-specific drug delivery system (CDDS) has been developing as one of the site-specific drug delivery systems. This delivery system, by means of combination of one or more controlled release mechanisms, hardly releases drug in the upper part of the gastrointestinal (GI) tract, but rapidly releases drug in the colon following oral administration. The necessity and advantage of CDDS have been well recognized and reviewed recently.\(^1,3\) In view of CDDS specifically delivering drug to the colon, a lot of benefits would be acquired in terms of improving safety and reducing toxicity when treating local or systemic chronic diseases. First, as for treating localized colonic diseases, i.e. ulcerative colitis, Crohn’s disease and constipation, the optimal drug delivery system, such as CDDS, should selectively deliver drug to the colon, but not to the upper GI tract.\(^1\) For this reason, the drug concentration was significantly less in the upper GI tract, while increased considerably in the colon, resulting in alleviated GI side effects. Second, the colon is referred to as the optimal absorption site for protein and polypeptide after oral administration, because of the existence of relatively low proteolytic enzyme activities and quite long transit time in the colon.\(^2,3\) CDDS could provide reliable protection against GI enzymatic degradation by releasing the polypeptide and protein nearly unchanged and fully efficacious in the preferred colon, thereafter resulting in remarkably increased bioavailability for protein and polypeptide. Finally, CDDS would be advantageous when a delay in absorption is desirable from a therapeutical point of view, as for the treatment of diseases that have peak symptoms in the early morning and that exhibit circadian rhythms, such as nocturnal asthma, angina and rheumatoid arthritis.\(^4,5\)

Polysaccharides, the polymer of monosaccharide retains their integrity because they are resistant to the digestive action of gastrointestinal enzymes. The matrices of polysaccharides are assumed to remain intact in the physiological environment of stomach and small intestine but once they reach in the colon, they are acted upon by the bacterial polysaccharides and results in the degradation of the matrices. A large number of polysaccharides such as amylose, guar gum, pectin, chitosan, inulin, cycloexetrins, chondroitin sulphate, dextrins, dextrin and locust bean gum have been investigated for their use in colon targeted drug delivery systems. The most important fact in the development of polysaccharide derivatives for colon targeted drug delivery is the selection of a suitable biodegradable polysaccharide. As these polysaccharides are usually soluble in water, they must be made water insoluble by cross linking or hydrophobic derivatization. Very important is an optimal proportional of the hydrophobic and hydrophilic parts respectively and the number of free hydroxy groups in the polymeric molecule.\(^6,7,8\)

Mesalamine is used for the long-term maintenance therapy to prevent relapses of Crohn’s disease and ulcerative colitis. However, when mesalamine is administrated orally, a large amount of the drug is absorbed from the upper gastrointestinal tract, and causes systemic side effects. Free mesalamine undergoes rapid and nearly

**ABSTRACT**

The objective of the present study is to develop colon targeted drug delivery system by using Chitosan as a carrier for Mesalamine. Matrix tablets containing various excipients and Chitosan were prepared by wet granulation technique using different binder systems. The prepared tablets were evaluated for Hardness, Weight variation, Drug uniformity, Friability and In-vitro Drug release study. The surface of the device of best formulation was coated with Eudragit S100 to ensure that the device was more pH dependent and trigger the drug release only at higher pH. The final product is expected to have the advantage of being biodegradable and pH dependant. The matrix tablet containing Chitosan as a carrier and Hydroxypropyl methyl cellulose as binder was found to be suitable for targeting mesalamine for local action in the colon as compare to other matrix tablets containing different binders. Matrix tablets containing Chitosan released 97-99% of mesalamine in simulated colonic fluid. The stability study for prepared tablets at 40°C/75% relative humidity for three months showed no significant change in In-vitro drug release pattern. The results of in-vitro study indicate that matrix tablets containing Chitosan as carrier and Hydroxypropyl methyl cellulose as binder are most suitable to deliver the drug specifically in colonic region. The final formulation of mesalamine for colon-specific drug delivery gives pH, time and enzyme controlled release.

**Keywords:** Mesalamine, Matrix tablet, Chitosan, Hydroxypropyl methyl cellulose, Eudragit S100, Colon Specific drug delivery.
complete systemic absorption from the proximal intestine depending on concentration and local pH, followed by extensive metabolism. It is thus of tremendous importance to deliver mesalamine locally in order to reduce influences by systemic drug absorption causing adverse effects. Hence, selective delivery of mesalamine into the colon is required.

The aim of this study was to explore the feasibility of the colonic microorganism to develop CDDS by using Chitosan polysaccharide as a carrier and mesalamine as model drug.

Material and Methods

Materials
Mesalamine was gifted from Bec Chemicals Ltd., Ankleshav, Chitosan 220 from CIFT Cochin; Hydroxyl propyl methyl cellulose (HPMC) (K4M) was a gifted from Colorcon Ltd., India. Sodium CMC, PVP K-30, Microcrystalline cellulose, Talc and Magnesium stearate was obtained from National chemicals, Baroda.

Methods
Preparation of tablets
Sustained release matrix tablets were prepared by wet granulation method. All the ingredients were passed through sieve # 100, blended accurately, mixed and granulated using PVP K-30 in isopropyl alcohol, Sodium CMC (10 % aqueous solution) and HPMC K4M (10 % alcoholic solution) as granulating aid. The granules obtained were dried in oven at 50°C for 2 hours. After drying, granules passed through sieve # 16 to obtained uniform size granules. After sufficient lubrication matrix tablets were prepared using Cadmach single punch tablet machine (M/S. Cadmach Machinery Co. Pvt. Ltd, Ahmedabad) using 10 mm SC (Shallow concave) die and punch set. All the prepared Mesalamine tablets were stored in airtight container at room temperature for further study. Amount of drug in all formulation was kept constant. The Best tablet formulation was enteric coated with Eudragit S 100 to give pH dependent release. The surface of the device was coated with Eudragit S100 to ensure that the device was more pH dependent and trigger the drug release only at higher pH. The composition of various formulations is shown in Table 1.

Evaluation of formulated tablet
Tablet Hardness
The strength of tablet is expressed as tensile strength (Kg/cm²). The tablet crushing load, which is the force required to break a tablet into halves by compression. It was measured using a tablet hardness tester (Monsanto Tablet Hardness Tester, Mht -20, Campbell electronics).

Weight Variation Test
Weight variation test is done by weighing 20 tablets individually; calculating the average weight and comparing the individual tablet weight to the average.

Friability
Friability test is performed to assess the effect of friction and shocks, which may often cause tablet to chip, cap or break. Roche friabiliator (EF2, Eletrolab) was used for the Purpose. This device subjects a number of tablets to the combined effect of abrasion and shock by utilizing a plastic chamber that revolves at 25 rpm dropping the tablets at a distance of 6 inches with each revolution. Preweighed sample of tablets was placed in the friabiliator, which was then operated for 100 revolutions. Tablets were dusted and reweighed. Compressed tablets should not lose more than 1% of their weigh.

Thickness was measured by vernier caliper. (SV -03, E-Base measuring tools)

Content Uniformity
Twenty tablets of mesalamine were weighed and powdered. Crushed powder of tablets equivalent to 0.15 gm was weighed and dissolved in pH 7.4 Sorensen’s Phosphate buffer. Solution was filtered and diluted and drug content was analyzed spectrophotometrically at about 334.5 nm.

In vitro drug release study
The ability of the matrix tablets of Mesalamine to remain intact and to release the active ingredient in the physiological environment of stomach, small intestine and colon was assessed by conducting in vitro drug release studies under conditions mimicking mouth to colon. The drug release studies (n= 3) were carried out using USP dissolution Rate test apparatus at 100 rpm and 37 ± 0.5°C. 900 ml. of 0.1 M HCL was used as dissolution medium in the first two hrs of study as the average gastric emptying time was estimated as 2 h. 5 ml. of the dissolution medium was withdrawn after 2 hr to determine the drug release. The volume withdrawn with replaces with fresh media and was accounted during calculation of cumulative percentage drug release. The amount of drug release was analyzed by Double beam UV spectrophotometer (Lambda 2, Perkin–Elmer, USA) at maximum wavelengths of 301.5 nm. The dissolution media was replaced at the end of two hrs. with pH 7.4 Sorensen’s Phosphate buffer, 900 ml. and drug release study was continued for another 3 hr. (i.e. total 5 hrs) as the average small intestine transit time is about 3 hrs. As before, samples were withdrawn at regular time intervals and correspondingly replaced with fresh media. The amount of drug release was analyzed by Double beam UV spectrophotometer (Lambda 2, Perkin–Elmer, USA) at maximum wavelengths of 334.5 nm.

Preparation of Rat caecal content medium:
Before starting the experiments on animals, the experimental protocol was subjected to the scrutiny of the Institutional Animal Ethical Committee (IAEC), and was approved by the same in time. The susceptibility of the matrix tablet to the enzymatic action of colonic bacteria was assessed by performing the drug release in medium contacting rat caecal content. Caecal material was collected from male albino rats weighing 150 -200 gm. Maintained on normal diet, but the caecal enzme production was induced by giving orally 1ml. of 2% w/v dispersion of pectin for 7 days (administered directly into the stomach using Teflon tubing) Thirty minutes before the commencement of drug release studies, four rats were killed and abdomen were opened, the caecai was isolated, ligated at both ends, cut loose and immediately transferred into pH 6.8 Phosphate Buffered Saline (PBS), previously bubbled with carbon dioxide (CO₂). The caecal bags were opened and their contents were individually weighed, pooled and then suspended in PBS to give a final caecal dilution of 4% w/v. The dissolution study was continued in 100 ml. of the above made rat caecal media after the 5th hr. this is done with slight modification in the experimental set up of the USP dissolution rat test apparatus. A beaker of 150 ml capacity containing 100 ml of PBS (pH 6.8) with rat caecal content was placed suitably in the dissolution vessel having water maintained at 37 ± 0.5°C which in turn was kept in the water bath of the apparatus. The study was continued from 5 hr to 24 hr and samples were withdrawn at regular intervals for analysis and each time replaced with fresh PBS
media containing rat caecal material bubbled with CO₂. The withdrawn samples were diluted with PBS and centrifuged. The supernatant was filtered through a bacteria proof filter and filtrate was analyzed for Mesalamine content using a Double beam UV spectrophotometer (Lambda 2, Perkin–Elmer, USA) at maximum wavelengths of 299 nm.

**Drug Release Kinetics**

To study the release kinetics, data obtained from in vitro drug release studies were plotted in various kinetic models: zero order (Equation 1) as cumulative amount of drug released vs. time, first order (Equation 2) as log cumulative percentage of drug remaining vs. time, and Higuchi’s model (Equation 3) as cumulative percentage of drug released vs. square root of time.

\[
C + K_0 t \\
\text{Where } K_0 \text{ is the zero-order rate constant expressed in units of concentration/time and } t \text{ is the time in hours. A graph of concentration vs. time would yield a straight line with a slope equal to } K_0, \text{ and intercept the origin of the axes.}
\]

\[
\log C + \log C_0 - kt/2:303 \\
\text{Where } C_0 \text{ is the initial concentration of drug, } k \text{ is the first order constant, and } t \text{ is the time. (Bourne 1963)}
\]

\[
Q + Kt^{0.5} \\
\text{Where } K \text{ is the constant reflecting the design variables of the system and } t \text{ is the time in hours. Hence, drug release rate is proportional to the reciprocal of the square root of time.}
\]

**Mechanism of Drug Release**

To evaluate the mechanism of drug release from Matrix tablet, data for the first 60% of drug release were plotted in Korsmeyer el al.’s equation (Equation 4) as log cumulative percentage of drug released vs. log time, and the exponent n was calculated through the slope of the straight line.

\[
M/M_0 = K t^n \\
\text{where } M/M_0 \text{ is the fractional solute release, } t \text{ is the release time, } K \text{ is a kinetic constant characteristic of the drug/polymer system, and } n \text{ is an exponent that characterizes the mechanism of release of tracers.}
\]

For cylindrical matrix tablets, if the exponent n = 0.45, then the drug release mechanism is Fickian diffusion, and if 0.45 < n < 0.89, then it is non-Fickian or anomalous diffusion. An exponent value of 0.89 is indicative of Case-II Transport or typical zero-order release mechanism. If 0.45 < n < 0.89, then it is non-Fickian or anomalous diffusion. An exponent value of 0.89 is indicative of Case-II Transport or typical zero-order release mechanism.

**Stability study**

Best formulation was (F9) exposed to three months stability study at 40°C/75% RH. These samples then again evaluated for drug release study.

**Results and Discussion**

Tablets were prepared using wet granulation technique. Tablets were obtained of uniform weight due to uniform die fill, with acceptable weight variation as per pharmacopeial specification. The drug content found in the range of 98.36-102.35% (acceptable limit) and the hardness of the tablet was found between 4.0 - 6.03 kg/cm². The tablet thickness was found to be around 3.0 mm, friability of tablet was found below 1% indicating good mechanical resistance. The formulated matrix tablets have content uniformity 95.68 to 104.63 %, (Table 2).

The in vitro release of mesalamine for all the nine batches are shown in Figure 1. The result showed that Sodium CMC and PVP K-30 binder along with Chitosan polysaccharides could release the mesalamine in gastric and intestinal fluid. In case of Sodium CMC, up 60% drug was released in first 5 hrs, but if the concentration of Chitosan was increased (batch F3) the drug release was decreased up to 43%. While in case of PVP K-30 as a binder, the batch F6 (highest Chitosan concentration) was released 36%. So, it was decided that Sodium CMC and PVP K-30 along with polysaccharide Chitosan could release the mesalamine in gastric and intestinal fluid. It was concluded that drug release from tablets up to 24 hrs. depend on amount of Chitosan Polysaccharides. Chitosan is a novel mucoadhesive polymer. It is totally degraded by colonic bacteria but is not digested in the upper GI tract. These polysaccharides remain intact in the physiological environment of the stomach and the small intestine, but are degraded by the bacterial inhabitants of the human colon. So it was concluded that chitosan is used in matrix forming tablet for colon targeting. Drug was released only when dosage form entered in colon where colonic bacteria degrade the polysaccharides. So, at higher level of chitosan, drug was released after 8 hrs.

But results showed that HPMC K4M is a good binder for matrix tablet formulations. The In-vitro release study of HPMC containing tablets showed that, at highest chitosan concentration (Batch F9), only 12% release of mesalamine occurs. Drug release studies show that F9 shows good release behavior in colon and restricts release in stomach and intestine. This study confirms that chitosan can act as good carrier in the form of matrix tablet for mesalamine to deliver it in colon specifically by using HPMC as binder. HPMC is a swellable polymer that shows time dependent release profile when the tablet comes into contact with liquids. HPMC K4M has viscosity of 3000-5600 cps and was used for matrix formulation.

From In-vitro release study, it was concluded that higher percentage of chitosan and HPMC retard the drug release, but drug release from batch F9 was found to be satisfactory because, from this batch drug release was found to be less than 15% in 5 hrs. And total drug was released in 24 hrs. Which was controlled by enteric coating so, batch F9 was promising batch and further enteric coating and stability study was performed on F9 batch. (Figure 2)

The zero-order rate describes the systems where the drug release rate is independent of its concentration. The first order describes the release from systems where the release rate is concentration dependent. Higuchi’s model describes the release of drugs from an insoluble matrix as a square root of a time-dependent process based on Fickian diffusion. The release constant was calculated from the slope of the appropriate plots, and the regression coefficient (r²) was determined (Table 3). It was found that the in vitro drug release of batch F9 was best explained by Higuchi’s equation, as the plots showed the highest linearity (r² = 0.9925), followed by zero order (r² = 0.980). This explains why the drug diffuses at a comparatively slower rate as the distance for diffusion increases, which is referred to as square root kinetics (or Higuchi’s kinetics). However, drug release was also found to be very close to zero-order kinetics, indicating that the concentration was nearly independent of drug release. The dissolution data were also plotted in accordance with the Hixson-Crowell cube root law. The applicability of the formulation to the equation indicated a change in surface area and diameter of the tablets with the progressive dissolution of the matrix as a function of time.

The corresponding plot (log cumulative percent drug re23.
Table 1: Composition of different Chitosan matrix tablets of mesalamine

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Ingredients</th>
<th>Quantity/tablet (mg)</th>
<th>Formulation codes</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>F1</td>
<td>F2</td>
</tr>
<tr>
<td>1</td>
<td>Mesalamine</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>2</td>
<td>Microcrystalline Cellulose</td>
<td>340</td>
<td>280</td>
</tr>
<tr>
<td>3</td>
<td>Chitosan</td>
<td>50</td>
<td>75</td>
</tr>
<tr>
<td>4</td>
<td>Sodium CMC (10% aq. solution)</td>
<td>05</td>
<td>10</td>
</tr>
<tr>
<td>5</td>
<td>Alcohol P.V.P K-30</td>
<td>——</td>
<td>——</td>
</tr>
<tr>
<td>6</td>
<td>Hydroxypropyl methyl cellulose K4M</td>
<td>——</td>
<td>——</td>
</tr>
<tr>
<td>7</td>
<td>Magnesium Stearate</td>
<td>2</td>
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</tr>
<tr>
<td>8</td>
<td>Talc</td>
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</tr>
<tr>
<td></td>
<td>Total weight of tablet</td>
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</tr>
</tbody>
</table>

Table 2: Evaluation Parameters of different CHITOSAN matrix tablets of mesalamine

<table>
<thead>
<tr>
<th>Formulation code</th>
<th>Average weight of Tablets (mg)</th>
<th>Average Hardness of Tablets (kg/cm²)</th>
<th>Friability of Tablets (%)</th>
<th>Average Thickness of Tablet (mm)</th>
<th>Assay (% Drug Content)</th>
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<tbody>
<tr>
<td>F1</td>
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<td>3.1</td>
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<td>F2</td>
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<td>0.42</td>
<td>3.2</td>
<td>102.35</td>
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<tr>
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<td>4.3</td>
<td>0.40</td>
<td>3.5</td>
<td>99.65</td>
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<tr>
<td>F4</td>
<td>496</td>
<td>5.5</td>
<td>0.39</td>
<td>3.0</td>
<td>98.36</td>
</tr>
<tr>
<td>F5</td>
<td>492</td>
<td>6.2</td>
<td>0.36</td>
<td>3.2</td>
<td>98.67</td>
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<tr>
<td>F6</td>
<td>493</td>
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<td>0.39</td>
<td>3.1</td>
<td>99.26</td>
</tr>
<tr>
<td>F7</td>
<td>503</td>
<td>5.3</td>
<td>0.23</td>
<td>3.2</td>
<td>100.36</td>
</tr>
<tr>
<td>F8</td>
<td>502</td>
<td>5.8</td>
<td>0.20</td>
<td>3.0</td>
<td>101.36</td>
</tr>
<tr>
<td>F9</td>
<td>490</td>
<td>6.3</td>
<td>0.16</td>
<td>3.1</td>
<td>99.64</td>
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Table 3 - Release of Kinetics of Batch F6

<table>
<thead>
<tr>
<th>Zero Order</th>
<th>First Order</th>
<th>Higuchi</th>
<th>Korsmeyer-Peppas</th>
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<td>r²</td>
<td>K₄(h⁻¹)</td>
<td>r²</td>
<td>K₁(h⁻¹)</td>
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<tr>
<td>0.980</td>
<td>4.314</td>
<td>0.731</td>
<td>0.082</td>
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</table>

Fig. I - % Drug release pattern of different CHITOSAN matrix tablets of mesalamine.

Fig. II - % Drug release pattern of Enteric coated and Stability sample of F9

Conclusion

The results obtained indicate that Chitosan could be useful as matrix system for controlled drug delivery and various polymers could be used to modulate the drug release from the matrix tablet depending on the need. The matrix tablets were passed in different evaluation test, content uniformity and in vitro drug release study. Drug release profile was evaluated in simulated gastric, intestinal fluid and simulated colonic fluid. Best formulation was F9 on the basis drug release pro-
file in simulated gastric, intestinal and colonic fluid. The matrix tablet containing chitosan as a carrier and HPMC as binder was found to be suitable for targeting mesalamine for local action in the colon as compared to other matrix tablets containing different binders. The surface of the device was coated with Eudragit S100 to ensure that the device was more pH dependent and trigger the drug release only at higher pH. The final product is expected to have the advantage of being biodegradable and pH dependent. Matrix tablets containing chitosan released 97-99% of mesalamine in simulated colonic fluid. Drug release kinetics indicated that drug release was best explained by Higuchi’s equation, as these plots showed the highest linearity ($r^2 = 0.9949$), but a close relationship was also noted with zero-order kinetics ($r^2 = 0.9850$). Korsmeyer’s plots indicated an $n$ value of 0.60, which was indicative of an anomalous diffusion mechanism or diffusion coupled with erosion; hence, the drug release was controlled by more than one process. Tablets containing chitosan showed no change in physical appearance and dissolution profile upon storage at 40°C/75% relative humidity for three months. The results clearly demonstrate that chitosan has the potentials for drug targeting to the colon.

**References**


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